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A comparative study of the marine ecoregions of the southern Iberian Peninsula, as identified from different coastal habitats

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ABSTRACT

EC Directive 2000/60 of the European Parliament and of the Council of Europe provides for the need to establish ecological regions along European coasts. The present article outlines research carried out in various coastal marine ecosystems as a contribution to the characterization of these ecoregions of the coast of Andalusia (southern Spain). Sampling was undertaken along the coastline in the intertidal zone and sublittoral sediments. The various systems studied all gave very similar results with regard to the ecoregions identified, which is indicative of their robustness. These results consistently identified a main Mediterranean region, which could subsequently be subdivided into two, as well as three main ecoregions in the Atlantic: one from the Straits of Gibraltar to the Bay of Cadiz, and the other two along the Huelva coastline, which are clearly affected by the effluents of major river systems such as the Guadalquivir, Odiel and Tinto. Our results suggest that similar structuring factors are operating within these systems.

Keywords: EC Directive 2000/60, coastal management, conservation, benthic communities, soft bottom, intertidal.

RESUMEN

Estudio comparado de las ecorregiones marinas del sur de la península Ibérica identificadas a partir de diferentes hábitats

La Directiva 2000/60 CE del Parlamento Europeo y del Consejo Europeo contempla la necesidad de establecer regiones ecológicas a lo largo del litoral europeo. Con el objetivo de contribuir a la delimitación y caracterización de estas ecorregiones se ha realizado este estudio de varios ecosistemas marinos centrado en el litoral andaluz (sur de España). Los muestreos se realizaron en la franja intermareal rocosa y en sedimentos infralitorales. Los sistemas estudiados dieron resultados similares con respecto a la definición de las distintas ecorregiones. Así, se identificó una gran área mediterránea, que podría dividirse en dos, y otra atlántica, que se divide en tres ecorregiones: una comprende desde el estrecho de Gibraltar hasta la bahía de Cádiz y otras dos emplazadas en el litoral de Huelva caracterizadas por la presencia de importantes efluentes fluviales como el Guadalquivir, el Odiel y el Tinto. Los resultados sugieren que existen factores estructurales similares en estos sistemas.

Palabras clave: *Directiva 2000/60 CE, gestión del litoral, conservación, comunidades bentónicas, sedimento, intermareal.*

INTRODUCTION

The EC Directive 2000/60 of the European Parliament and of the Council establishes a framework for community action in the field of water policy (European Union, 2000) and provides for the need to establish ecological regions along the European coasts which will make it possible to compare biological control data.

The identification of various regions of similar taxonomic composition to aid ecological research and conservation efforts has a long history (Herbertson, 1905; Thornthwaite, 1933; Pitelka, 1941; Holdridge, 1947). Such ecoregions are still one of the main tools used by conservation ecologists, and many variants of the concept have been developed (e.g. Walter and Box, 1976; Bailey, 1983), although the subjective approach taken by a number of researchers in delineating their ecoregions has led to there being as many sets of ecoregions as there are researchers (Hargrove and Luxmoore, 1998).

The definition of ecoregions via the use of more empirical and objective data analyses, such as multivariate clustering techniques (e.g. Omi, Wensel and Murphy, 1979), is an alternative to maps based on expert opinion. This is the approach used in the present study.

In marine research circles, the establishment of ecoregions has gained increasing popularity, particularly with regard to the creation of readily identifiable biogeographic entities that are amenable to conservation efforts, such as legislation (Belbin, 1983; Belfiore, 2000; Snoussi and Tabet-Aoul, 2000; Roff , Taylor and Laughren, 2003).

The composition of benthic communities is particularly useful as a baseline for ecological monitoring, and thus lends itself to the establishment of ecoregions because of the sessile nature of most of the organisms present, so that the community reflects their history. The need to rapidly establish biogeographically robust ecoregions is necessarily determined by the required data resolution and associated logistics (e.g. sampling, sorting, and identifying), which can be especially complex when sampling in the marine environment. Thus, any development that would simplify and thus expedite the establishment of marine ecoregions is welcome.

In the present paper, we compare the marine ecoregions identified via two independent studies involving very different target ecosystems and sampling methodologies, but both based on identification and characterization of benthic macrofaunal communities. The first study, with financial and technical support from the Andalusian Regional Government's Department of the Environment (Consejería de Medio Ambiente), was based on sediment benthic grab-samples collected during a series of cruises off the southern Iberian Peninsula, which were subsequently processed in the laboratory. The second study (carried out along the same biogeographical area) was conducted on rocky shores in situ, using belt transects and data collection. The degree of concordance between the results obtained would then establish whether different marine benthic systems produce similar biogeographic groups, thus establishing (or not) the generality of the observed ecoregions.

MATERIALS AND METHODS

For both studies, macrofauna are defined as organisms > 1 mm in size.

Method 1: Sandy-bottom benthic survey

A total of 85 points were sampled, spanning the coast of Andalusia from Huelva in the west to Almeria in the east. Sampling points were located with a GPS, and an echo-sounder was used to determine depth. Figure 1 shows the geographical extent of the study.

Ship-mounted Van Veen dredges, with a grab area of 0.05 m^2 and weighing approximately 8 kg, were used to collect the sediment samples at each site. The samples were subsequently bagged for later transfer to the laboratory. This type of dredge makes it possible to sample rectangular areas, and, independently of the sediment type, will penetrate to a minimum depth of 10 cm, thus ensuring the collection of the sediment fraction that contains the majority of the fauna of interest to this study.

Five replicate samples were taken at each site, giving a total sampled area of 0.25 m^2. Various authors have agreed that a sample surface area of 0.1 m^2 is the minimum required to adequately assess taxonomic diversity in sediment (Gentil and Dauvin, 1988; Ortiz, 2002), so the final aggregate sample size per site was considered to be representative of the zone under study. Approximately half

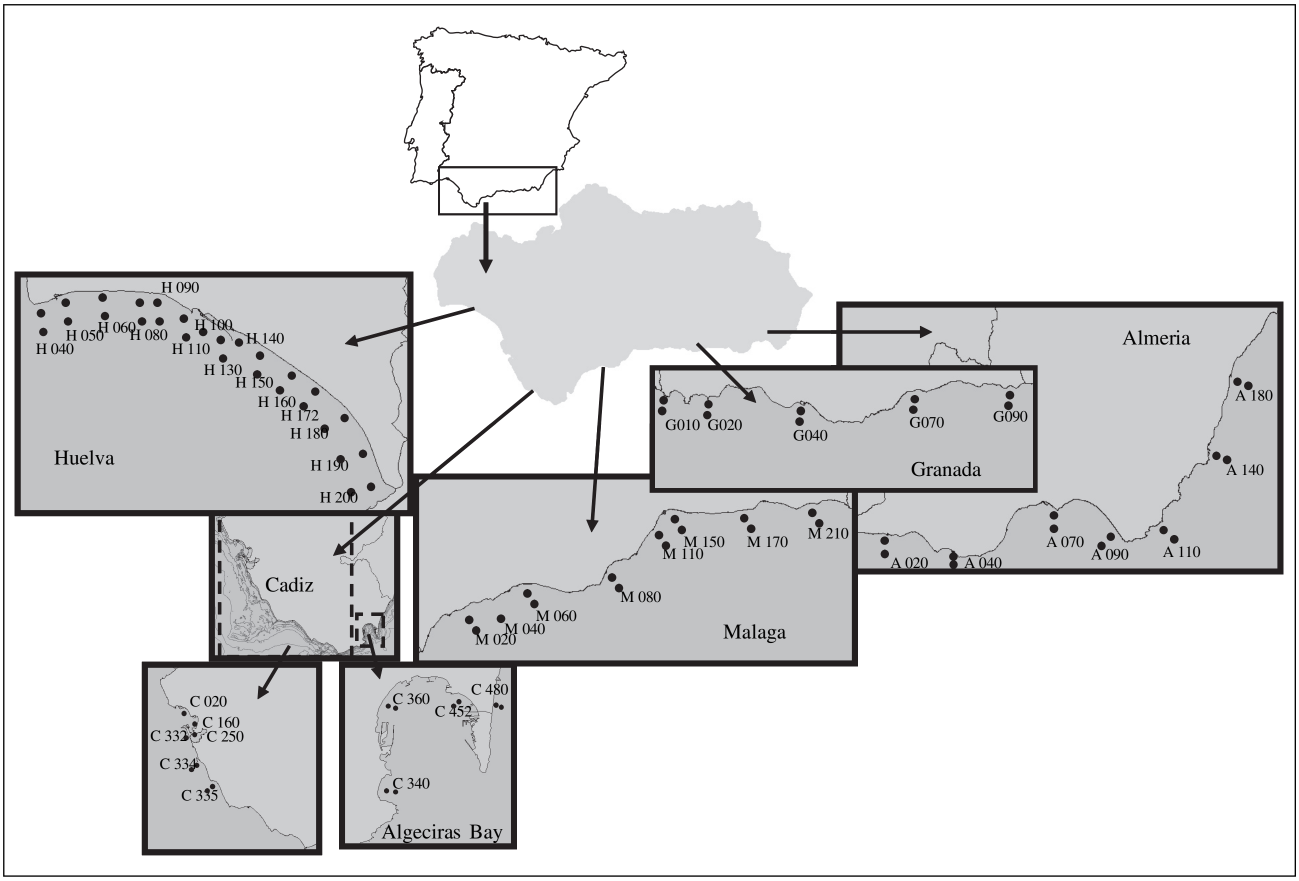


Figure 1. Location of the five main areas and the 85 points sampled for this study

of the samples were taken at a depth of around 10 m, and the other half at 20 m. These would allow for further comparisons between the ecoregions established at each given depth, as well as producing ecoregions based on aggregated data.

Although we defined macrofauna as organisms > 1 mm in size, sediment samples were sieved using a 0.5 mm mesh. This technique would retain some meiofauna (organisms < 0.5 mm), but would also allow the retention of juvenile forms of macrobenthic species, an important component of the community structure which in turn provided us with important information on the environmental quality of the site. Moreover it ensured that vermiform organisms (which would most likely pass through larger mesh sizes) could be trapped, thus enhancing both the qualitative and quantitative data obtained. After sieving, the samples were fixed using 10 % formaldehyde solution and stained using Bengal rose, to facilitate the visual extraction of individuals from the surrounding sediment. Following separation, organisms were identified, counted and conserved.

Taxonomic identification was to family level. The use of taxonomic levels higher than species has been in use in the ecological sciences for a number of years and by various authors (Herman and Heip, 1988; Warwick, 1988a,b; Ferraro and Cole, 1990; Gray *et al.*, 1990; Warwick *et al.*, 1990; Warwick and Clarke, 1993; Balmford, Green and Murray, 1996; Balmford, Jayasuriya and Green, 1996). Even in cases where environmental perturbations are relatively weak and are undetectable, using univariate methods (such as diversity indices) at species level, multivariate analyses at higher taxonomic levels has managed to detect these effects, and in some cases has made such results more evident, since it reduces the amount of noise generated by species-level analyses, especially where many of these species are adapted to narrow ranges of environmental conditions (Warwick, 1993). For each of our samples, the taxonomic families and number of individuals were noted and used in the subsequent statistical analyses. A further analysis of the whole-site data was also carried out after removing all species which contributed < 1 % to the total abundance to establish whether the results obtained were disproportionately affected by rare taxa.

Method 2: Rocky intertidal survey

A systematic method based on a fixed belt transect (Eberhardt, 1978; Jones *et al.*, 1980; Fa, 1998) was selected as the sampling method at each site for the quantitative assessment of changes in abundance and distribution of littoral organisms. A total of 20 sites were sampled, spanning the southern Iberian Peninsula from Vila Nova de Milfontes in Portugal to La Manga del Mar Menor in southeast Spain, covering a total distance of approximately 1 500 km (figure 2). Each transect was also selected to be as consistent as possible throughout all sites (in terms of general profile, slope and topography). This was done so as to minimise the effects of topographical heterogeneity and allow the data from all sites to be more favourably cross-compared (Fa, 1998).

At each site a generalised shore search was undertaken in order to both select the most appropriate location for the transect, and also to ensure that it was as representative as possible of the area being studied. Once the transect area had been chosen, the transect itself was measured and marked off. Vertical heights (from MLW) were marked off with a penknife and later correlated to the relevant chart datum (information supplied by the Gibraltar Meteorological Office, Instituto de Meteorología, Madrid and the Admiralty Tide Tables). Details of the technique used to measure vertical heights can be found in Fa (1998) and Fa *et al.* (2002).

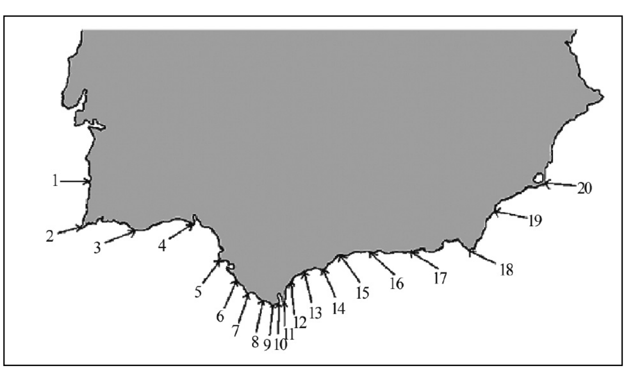


Figure 2. Location of the twenty sampling points for the rocky shore study

Sampling points (stations) were marked off at 25 cm intervals. This narrow vertical height was selected in order to assess the very compressed existing zonation patterns, particularly in the Mediterranean. This process was continued until a vertical height of 3 m had been achieved, providing 12 stations for the littoral zone. A rectangular fixed-area 0.25 m^2 (1 m × 0.25 m) quadrat, placed vertically against the substrate and parallel to the air-water interface, was used to sample the macrofaunal assemblages. A rectangular quadrat was used, as it helped to maintain the area being sampled as environmentally homogeneous as possible. This was needed, because the small tidal amplitudes encountered –particularly in the Mediterranean– meant that the recognised littoral zones showed extreme vertical compression (Fa, 2000; Fa and Sheader, 2000). Sampling with a square quadrat would aggregate distinct assemblages and have produced a diffuse zonation pattern for the shore. Within a shore level, a rectangular quadrat also increases the probability of bridging the various vegetational mosaic components and hence reduces the variance of the data (Dalby, 1987).

A variant of Pielou's pooled quadrat method (1975) was used to assess minimum quadrat size. With this method, species diversity is continually recalculated as the sample size is steadily increased, and the point where the curve levels off is taken to be the minimum quadrat size required in order to adequately sample the assemblages. Details of this method can be found in Fa (1998).

Within each of these stations, a census of the species richness and the number of individuals encountered was taken. Due to the mainly two-dimensional nature of the rocky littoral, it proved possible to make this assessment on site, with only a few of the smaller and more motile species proving difficult to capture and consequently only a rapid visual identification was possible, so that identification was only down to genera. In any case, it is important to note that in almost all the above cases, the species in question were rare and only encountered at a single location, making the degree of taxonomic resolution less important, as they essentially act as morphospecies. In other cases, identification was purposefully taken only to genera level, due to the difficulties of on-site separation of very similar co-existing species (e.g. *Mytilus edulis* Linnaeus, 1758 and *Mytilus galloprovincialis* Lamarck, 1819, whose morphs grade into each other, and consequently they were grouped as *Mytilus* spp.).

Statistical treatment

As entire sites were to be compared in this analysis, the values for each site were aggregated to give a single value for taxonomic richness and number of individuals at each site. The similarity between any two samples was calculated using the Bray-Curtis similarity index (Bray and Curtis, 1957), following data reduction using a root-root or fourth root transform $y = \sqrt[4]{x}$ (Field, Clarke and Warwick, 1982; Field *et al.*, 1977) to avoid the swamping of rarer species by superabundant ones. Following this transformation, triangular similarity matrices between each pair of samples were calculated and a dendrogram created by hierarchical agglomerative clustering with group-average linking (e.g. Clifford and Stephenson, 1975), with which the existence of groupings between stations based on their similarities was obtained.

The ordination technique used to explore these patterns was non-metric Multi-Dimensional Scaling (MDS) (Kruskal and Wish, 1978, for an introduction). This is an ordination technique based on similarities between samples. By employing an iterative algorithm that progressively arranges samples in multi-dimensional space in order to best preserve their inter-sample dissimilarities and consequently arrive at the best ordination of these, where the distance between points in the ordination is proportional to the similarity between them (Gauch, 1982; Smith, Bernstein and Cimberg, 1987), it essentially allows species to "tell their story" (Field *et al.*, 1982). The degree to which the final representation fits the data is measured by a coefficient of stress.

In addition, the contribution of different species to average between-group dissimilarity and within-group similarity was assessed using the Simper program (analysis of percentage similarity), part of the Plymouth Routines in Multivariate Ecological Research (primer) package. The values obtained for the ratio of mean dissimilarity (or similarity) to its standard deviation ($\overline{\delta}_1/\mathrm{SD}\ (\overline{\delta}_1)$) for each species helped to establish species that contribute to a groups' separation from others or to its defining characteristics respectively.

RESULTS

Figures 3-5 show the results obtained following cluster analyses for the benthic study of aggregate, 10 m and 20 m samples. It should be noted that although multiple clusters are identified, a generally consistent pattern emerges: a large group containing the Mediterranean sites, which can be divided into two subgroups, and smaller Atlantic groups containing the sites from the Straits to Cadiz and from Huelva westwards, with a distinct subgroup of sites that are subjected to a high degree of exposure to river effluents.

Three main groups were identified with aggregate data (figure 3): one containing the Mediterranean points and a part of the Atlantic coast; another comprising most of the Atlantic coast (Cadiz and Huelva); and the third, points influenced by the mouths of rivers, such as the Guadalquivir (Huelva), Palmones (Cadiz), and Guadalhorce (Malaga).

Moreover, seven groups are shown with 10 m samples (figure 4), and by deleting those sites with specific characteristics or influenced by river effluents, we could divide the coast into four main areas: from Almeria up to the eastern zone of Granada; from Motril (point G040) up to the Straits of Gibraltar; the Atlantic coast of Cadiz; and the coast of Huelva, which could be subdivided in a east zone up to the mouth of the Tinto and Odiel rivers and in a west zone up to the Portuguese border (Guadiana River).

The 20 m sample cluster also shows seven groups which could be reduced to three areas: Mediterranean coast, up to the Straits of Gibraltar; Atlantic coast of Cadiz and west zone of Huelva; and east zone of Huelva, from the Guadalquivir's mouth up to the Tinto and Odiel mouths.

The MDS plots for the aggregate samples show a clear gradient from the zone occupied by the majority of the stations in the Huelva area through to the zone occupied by the Almeria stations (figure 6). Stations from the Cadiz area occupy a transitional placement. When stations at 10 m and 20 m are analysed and plotted separately (figure 7), a tendency for shallower stations to exhibit a higher degree of discrimination between sites than the deeper ones becomes evident. This is consistent for each of the provinces surveyed.

Although the cluster (figure 8) and MDS (figure 9) for the aggregate sample, after removing species that contributed less than 1% to the dataset,

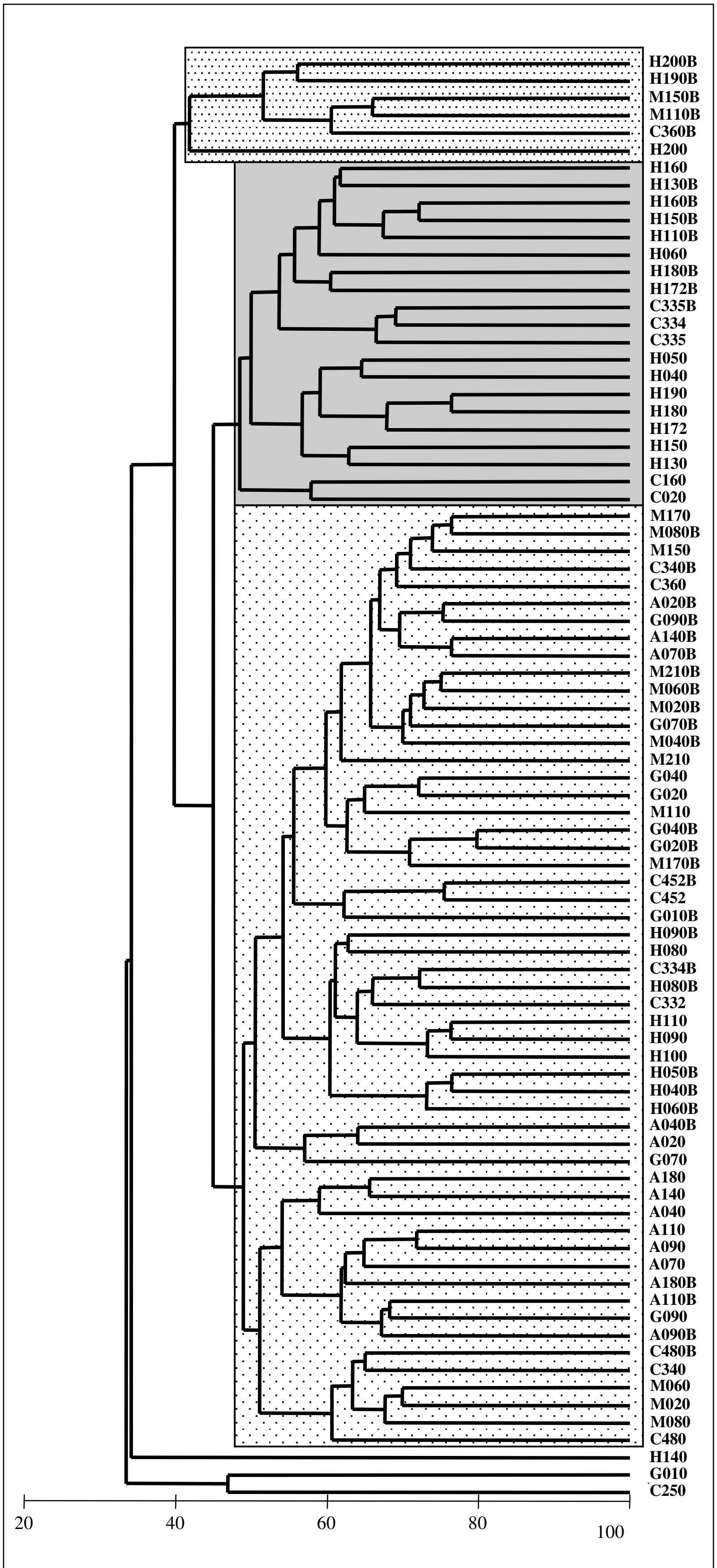


Figure 3. Cluster analysis for all (aggregate) benthic sites. Note that three main groups are identified

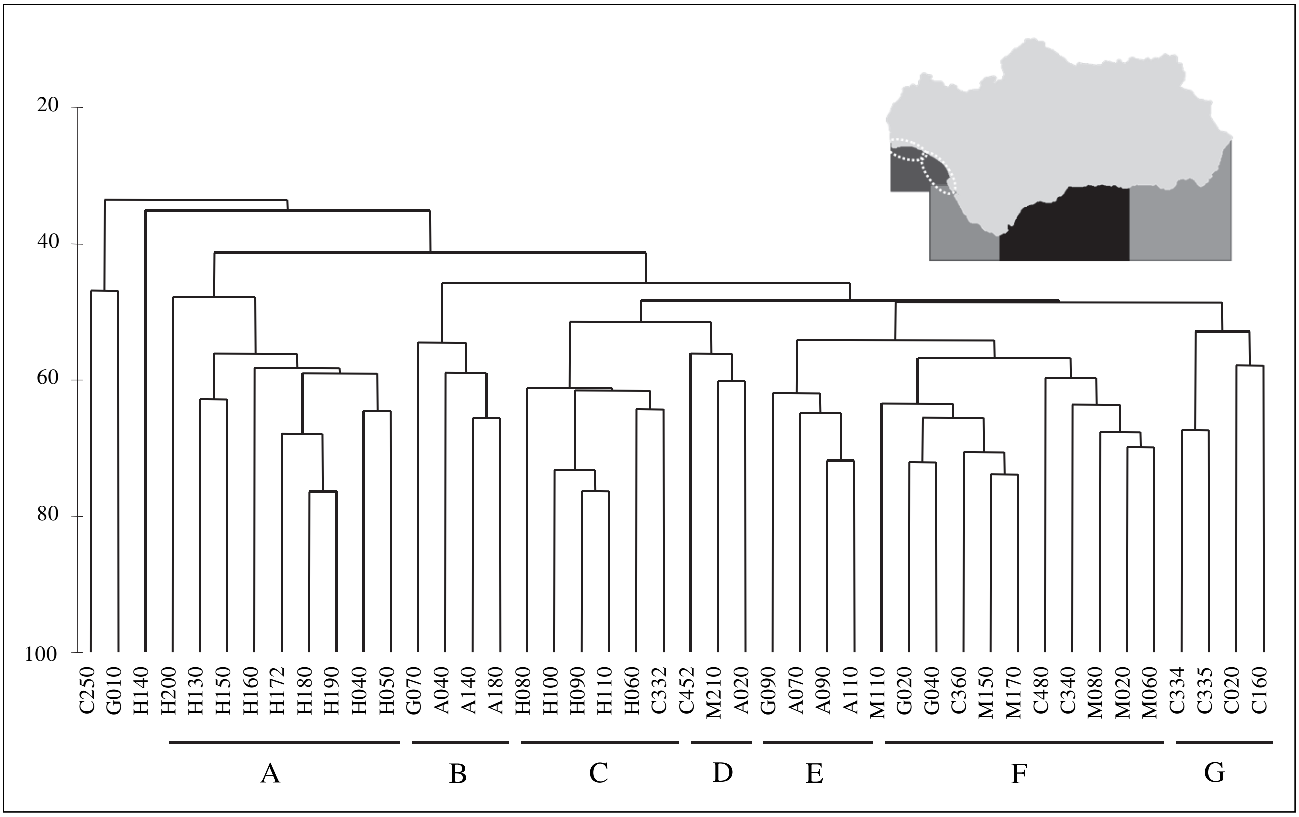


Figure 4. Cluster analysis for all 10-m benthic sites (note that the three main groups are still identified but the Mediterranean is now divided into two groups) and generalised graphical representation of the ecoregions as identified from the 10-m benthic samples

showed a large number of subgroups, in general five large groups emerge:

- A group that includes the vast majority of Mediterranean sites and which in turn can be subdivided into two large groups, one centred on the zone between Gibraltar and Estepona, and a second in the region of Cabo de Gata in Almeria.
- Atlantic coast of Cadiz (this zone shows certain similarity with the following group).
- A group which includes stations from west of Huelva up to the mouth of the Guadiana.
- A group which includes sites east of Huelva up to the mouths of the Tinto and Odiel Rivers.
- Sites that are heavily influenced by river outputs.

According to the results obtained for each of the analyses, figure 10 shows the final distribution of benthic ecoregions: 1) Mediterranean coast: subdivided into two, one from the Straits of Gibraltar to Motril (Granada), and another from this point to Almeria; 2) Atlantic coast of Cadiz; 3) east coast of Huelva: from Guadalquivir to the mouths of the Tinto and Odiel; 4) west coast of Huelva: from the Tinto and Odiel mouths to the Guadiana River (Portuguese border).

For the rocky shore study, the initial cluster analysis clearly identifies five groups, two of which form a larger Mediterranean group, a third covering the area from the Straits up to Cadiz, a fourth stretching west from Huelva, and a final group clearly defined by the impact of river effluents (figure 11).

Figure 12 is a MDS of the sites, and this again shows a clear gradient from Mediterranean to Atlantic sites. Table II shows the results of the Simper analysis. Although in the main, differences between clusters seem to be due to changes in abundance of certain species, there are some species that do appear to help to define the clusters due to their presence/absence. These species are highlighted in table I. The generalised distribution of ecoregions as defined by the rocky shore sites is shown in figure 13.

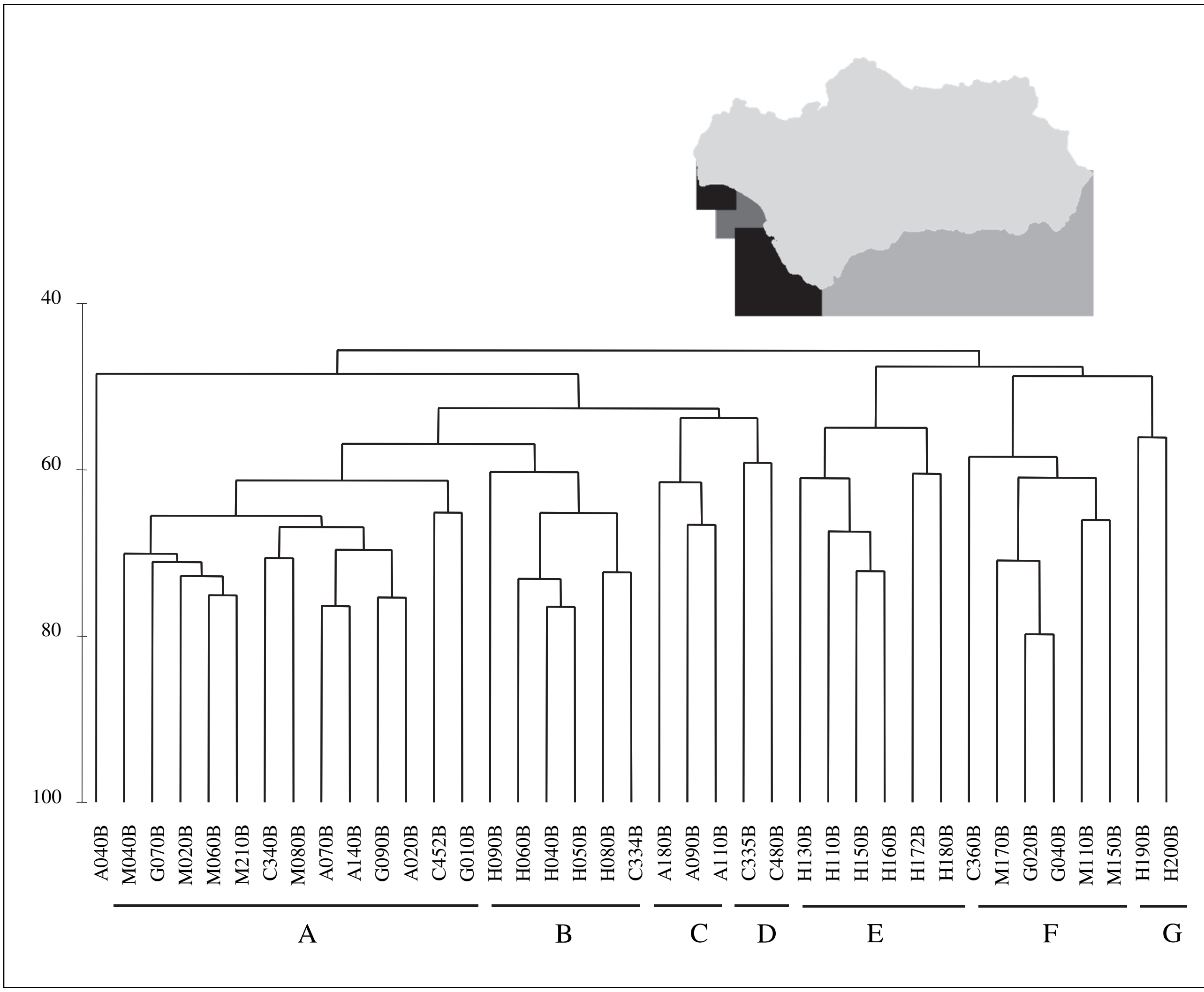


Figure 5. Cluster analysis for all 20-m benthic sites (note that the three main groups are again identified) and generalised graphic representation of the ecoregions as identified from the 20 m benthic samples

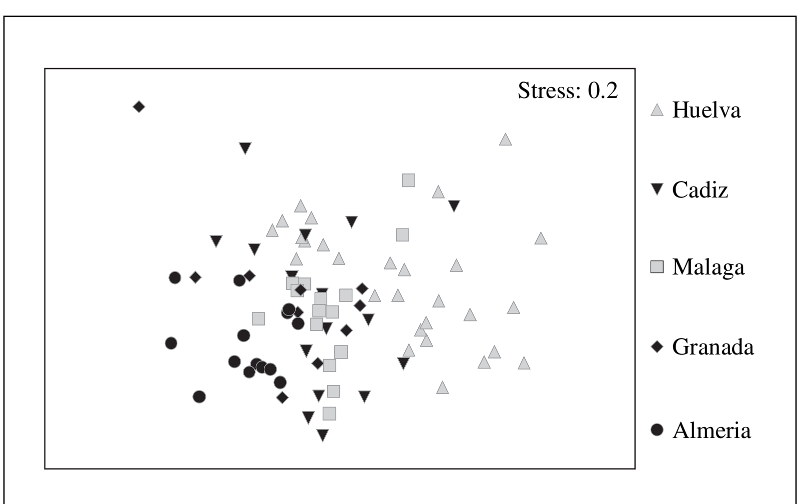
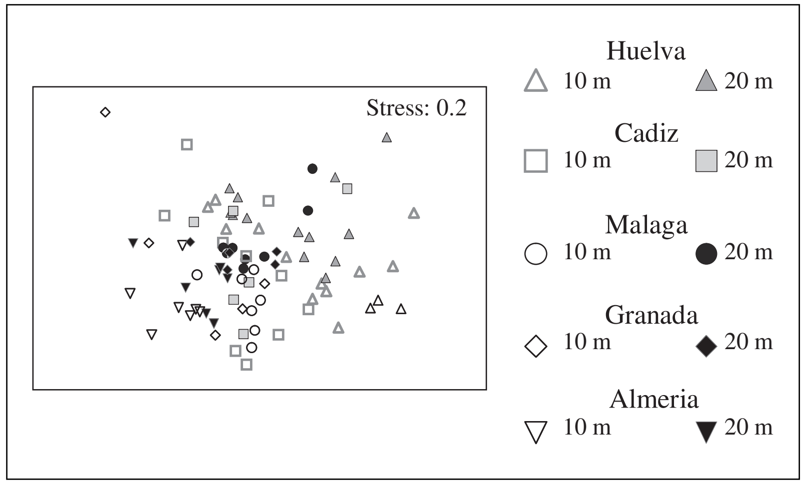


Figure 6. MDS of all aggregate benthic sites. Note the clear gradient from Atlantic through to Mediterranean sites

Figure 7. MDS of all 10-m and 20-m benthic sites. The gradient is clear for both samples, but shows a higher degree of discrimination in the shallower sites



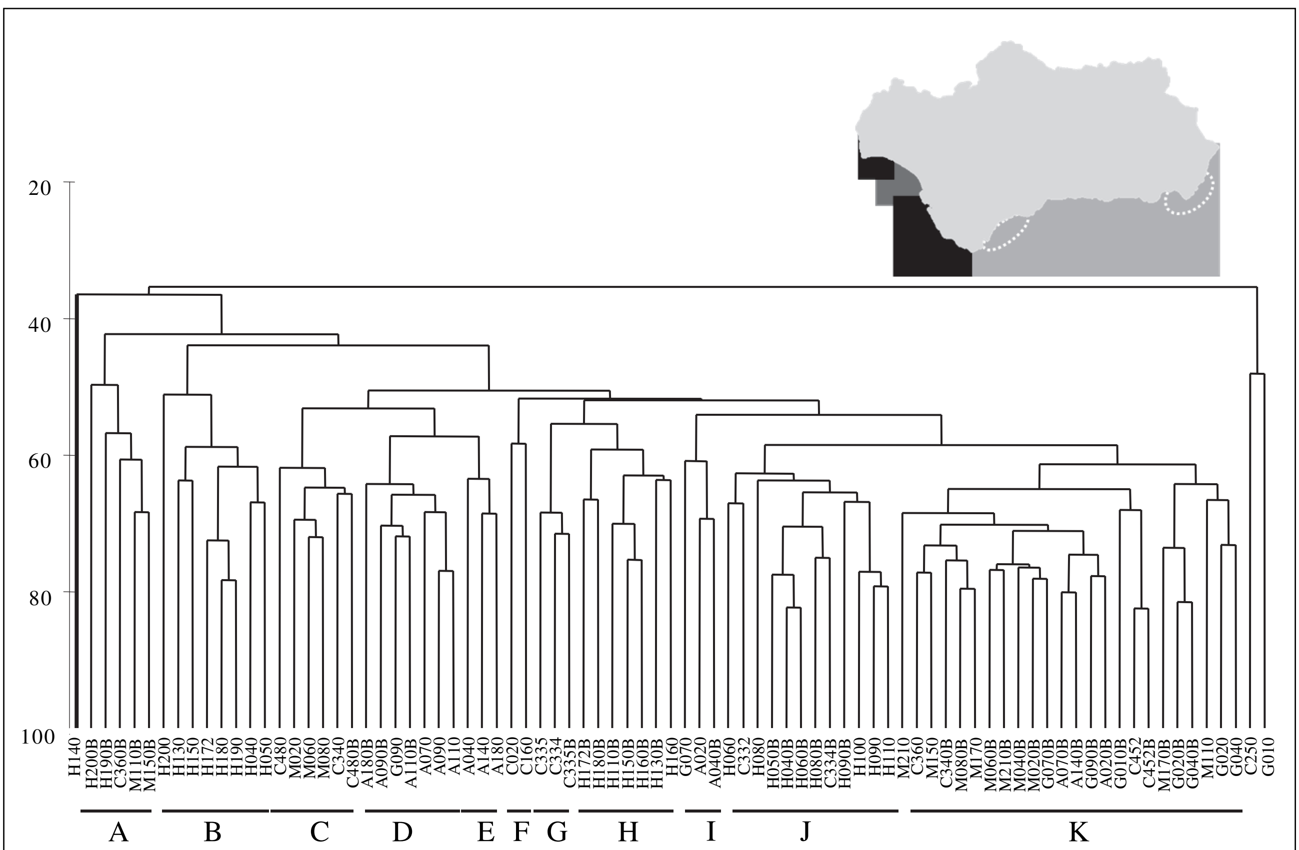


Figure 8. Cluster analysis for all (aggregate) benthic sites, after removing all taxa that contributed less than 1 % to the total abundance and generalised graphic representation of the ecoregions

DISCUSSION

This multivariate geographic clustering technique has several advantages over ecoregions drawn up by expert opinion. The groupings obtained are not only data-driven and empirical; moreover, they are not based on any particular taxonomic group.

Throughout all the analyses, the final graphic representations of the areas' ecoregions (figures 10

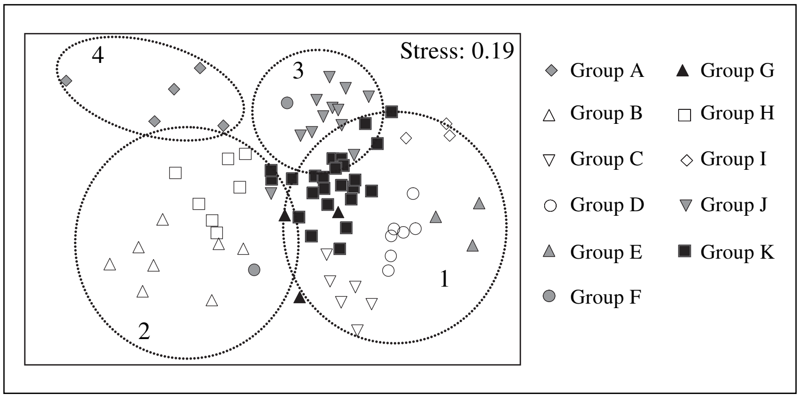


Figure 9. MDS of all (aggregate) benthic sites, after removing all taxa that contributed less than 1 % to the total abundance. Four main groups are identified, which are discussed further in the text

Table I. Results of a simper analysis for the rocky shore sites. Species are listed in order of their contribution ($\overline{\delta}_1$) to the average dissimilarity $\overline{\delta}_1$ (= 53.5) between the two groups, with a cut-off when the cumulative per cent contribution ($\Sigma\,\overline{\delta}_1$ %) to $\overline{\delta}_1$ reaches 80 %. Shaded species indicate a higher $\overline{\delta}_1/\mathrm{SD}\ (\overline{\delta}_1)$ ratio (> 1), marking them as particularly important contributors to between-group dissimilarity. Note that the species identified as accounting for over 25 % of the cumulative contribution (*Chthamalus* spp., *Mytilus* spp. and *L. punctata*) are all relatively abundant middle/upper shore species

Species	$\overline{y}_B$	$\overline{y}_A$	$\overline{\delta}_1$	SD($\overline{\delta}_1$)	$\overline{\delta}_1$/SD ($\overline{\delta}_1$)	$\overline{\delta}_1$ %	$\sum\overline{\delta}_1$ %
Chthamalus spp.	8 620.00	60 817.60	6.55	3.76	1.74	12.24	12.24
Mytilus spp.	379.80	9 099.80	4.76	2.57	1.85	8.89	21.13
Littorina punctata	231.27	0.00	2.91	1.57	1.85	5.44	26.58
Monodonta articulata	492.53	0.00	2.19	2.33	0.94	4.08	30.66
Chthamalus depressum	79.47	0.00	1.93	1.37	1.41	3.60	34.27
Balanus perforatus	49.73	358.60	1.82	1.43	1.27	3.40	37.67
Patella caerulea	58.53	146.40	1.43	0.94	1.52	2.67	40.34
Patella rustica	34.40	11.20	1.42	1.09	1.30	2.66	43.00
Gibbula umbilicalis	0.60	28.40	1.24	1.02	1.22	2.31	45.31
Scalpellum scalpellum	0.00	5.00	1.13	0.71	1.60	2.12	47.43
Patella aspera	5.93	15.80	0.94	0.75	1.25	1.76	49.19
Littorina saxatilis	4.27	10.40	0.94	0.78	1.21	1.75	50.94
Littorina littorea	0.00	15.00	0.91	0.78	1.17	1.70	52.65
Mercierella enigmatica	6.73	0.40	0.88	0.75	1.18	1.65	54.30
Anemonia sulcata	6.40	5.00	0.88	0.67	1.32	1.64	55.94
Syllidae/Nereidae	7.93	1.80	0.85	0.83	1.03	1.58	57.52
Pomatoceros triqueter	0.27	9.60	0.84	0.95	0.88	1.57	59.09
Carcinides maenas	7.53	3.40	0.79	0.67	1.18	1.48	60.57
Ligia spp.	1.80	3.80	0.78	0.72	1.09	1.45	62.02
Patella tarentina	8.13	17.60	0.75	0.61	1.23	1.40	63.42
Crassostrea angulata	0.00	2.80	0.72	0.90	0.80	1.34	64.76
Siphonaria pectinata	34.67	51.60	0.71	0.59	1.20	1.32	66.08
Diodora italica	3.13	0.00	0.71	0.76	0.94	1.32	67.40
Paracentrotus lividus	2.27	4.40	0.65	0.70	0.93	1.22	68.62
Actinia fragacea	0.40	2.00	0.65	0.60	1.09	1.22	69.84
Onchidella celtica	0.00	12.00	0.65	0.84	0.77	1.22	71.06
Gammaridae	142.40	145.60	0.63	0.47	1.34	1.19	72.25
Caprellidae	1.33	5.40	0.62	0.72	0.86	1.16	73.41
Rissoa spp.	2.73	0.00	0.60	0.77	0.78	1.13	74.54
Nucella lapillus	0.00	17.20	0.60	0.81	0.74	1.13	75.66
Chiton olivaceus	2.33	5.00	0.59	0.78	0.76	1.11	76.77
Cardita calyculata	0.00	10.20	0.59	0.75	0.79	1.11	77.87
Hydroides norvegica	0.00	4.80	0.53	0.65	0.81	0.99	78.87
Pachygrapsus marmoratus	6.33	15.40	0.50	0.37	1.36	0.93	79.79
Diodora apertura	1.80	0.00	0.48	0.62	0.77	0.89	80.68

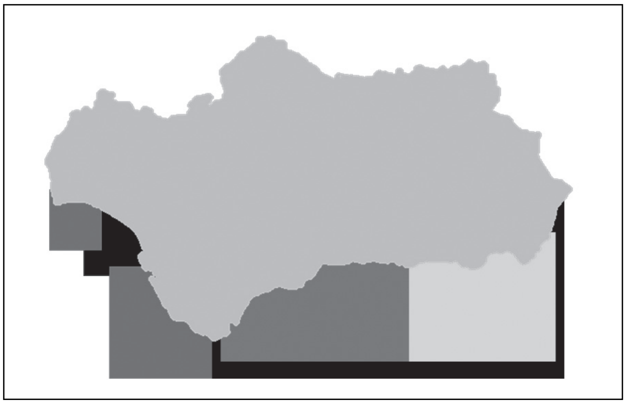


Figure 10. Generalised graphic representation of the ecoregions as identified from collating the various clusters obtained from the all the different analyses of benthic samples

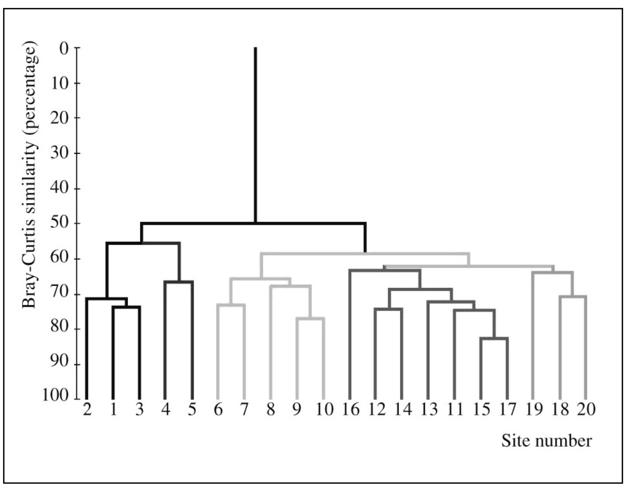


Figure 11. Cluster analysis for all (aggregate) rocky shore sites. Note that three main groups are identified, with the Mediterranean group being divisible into two subclusters

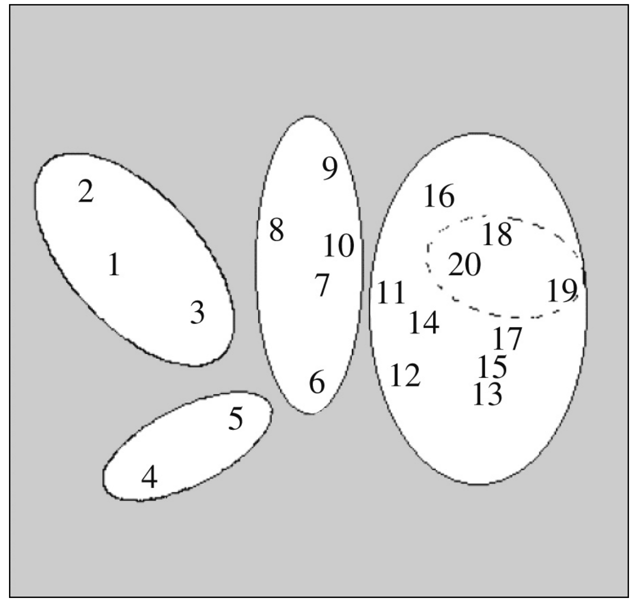


Figure 12. MDS of all aggregate rocky shore sites. Note again the clear gradient from the Atlantic through to the Mediterranean sites, and the overall similarity to the MDS obtained for benthic sites with regard to the grouping of sites into clusters

Figure 13. Generalised graphic representation of the ecoregions as identified from collating the various clusters obtained from the analysis of rocky shore sites. Note that it is virtually indistinguishable from that obtained for benthic sites (figure 10)

and 13) were essentially identical for both studies, so that a number of ecoregions have been identified consistently. These are a large Mediterranean group, encompassing the area from La Manga del Mar Menor (co-ordinates 37° 37.858' N, 00° 41.607' W) to the Straits of Gibraltar (36° 06.426' N, 05° 25.972' W), which is further divided into east and west sections around the area of Motril (36° 43.260' N, 03° 32.040' W) according to the soft-bottom communities and around the area of Adra (36° 45.896' N, 03° 06.657' W) as defined by the rocky shore sites; another group encompassing the area from the Straits of Gibraltar (Punta del Carnero/Getares), through the coastline of the Bay of Cadiz, through to the approximate location of Rota (36° 37.642' N, 06° 22.767' W); a smaller group from Rota up to the area of Huelva (37° 07.248' N, 06° 49.181' W), identified by the fact that it is heavily influenced by the effluents of major river systems such as the Guadalquivir, Odiel and Tinto; and a final group running west towards Portugal from Huelva (37° 08.613' N, 07° 23.251' W). The fact that these stem from different datasets suggests a robust, ecosystem-independent group of ecoregions, which may be indicative of similar, if not the same, generative processes. However, it must be remembered that the co-ordinates given are, at best, approximations, and that in any case the demarcations of these ecoregions are, by their very nature, diffuse and likely to show a degree of variability over both spatial and temporal scales. On the other hand, the different borders of the two Mediterranean sections established according to both ecosystems (Motril and Adra, respectively) suggest the possible presence of a transitional ecoregion in this area, although other studies will be necessary in order to elucidate this question.

It should be noted that, given the relatively simpler logistics involved and the similarity in results obtained, the rocky intertidal area would seem to be a useful candidate for the rapid establishment of coastal ecoregions (where suitable substrates exist).

In both studies, the main factors determining the clusters are changes in the overall abundance of certain species or taxa, but in both cases some species or taxa also emerge as possible candidates for a species-based delineation of ecoregions.

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